



NINHYDRIN REAGENT

- For in vitro use only -

Catalogue No. RN70

Our Ninhydrin Reagent is used to detect the end product, glycine, from rapid hippurate hydrolysis tests.

Ninhydrin reagent can only be used for rapid tests that contain **only** hippurate such as 1% Hippurate Broth devised by Hwang and Ederer (Dalynn TH65) and Hippurate Disks (Dalynn DH45); it is not suitable for hippurate tests that involve protein-containing growth media since ninhydrin may react with any free amino acids contained in the mediums.

The hydrolysis of hippurate liberates two major end products, glycine and benzoic acid. Ninhydrin reacts with glycine and through a complex reaction forms a purple end product that can be easily visualized.

Formulation per 100 mL

Ninhydrin..... 3.5 g
Acetone.....50.0 mL
Butanol50.0 mL

Recommended Procedure

Please refer to individual product technical sheets for detailed testing procedures:

DH45 Hippurate Disks
TH65 Hippurate Broth (0.5-mL)

Interpretation of Results

Positive (+): Deep purple-blue color
Hippurate hydrolyzed

Negative (-): Colorless (no change)
Hippurate not hydrolyzed

- Incubation beyond 30 minutes after the addition of Ninhydrin Reagent can result in false positive results
- Ninhydrin cannot be used to test for hippurate hydrolysis in protein-containing growth media such as Heart Infusion Broth or Todd-Hewitt Broth supplemented with hippurate. In such instances the proper reagent is Ferric Chloride Reagent (Dalynn RF40)

Quality Control

The following organisms are used to determine the performance of the product. A pure, overnight culture of each organism derived from a TSA plate is heavily inoculated into Hippurate Broth and incubated at 37°C for 2 hours. 0.2-mL of Ninhydrin Reagent is added to the broth, and after 10 minutes of incubation results are read.

Organism	Expected Results	
<i>Streptococcus agalactiae</i> ATCC 27956 (Group B)	+ve	Purple color
<i>Streptococcus pyogenes</i> ATCC 19615 (Group A)	-ve	No color change

Storage and Shelf Life

Our Ninhydrin Reagent should be stored at 4°C to 8°C. At this temperature it has a shelf life of 52 weeks from the date of manufacture.

References

1. Hwang M, Ederer GM. Rapid hippurate hydrolysis method for presumptive identification of group B streptococci. *J Clin Micro* 1975; 1:114-5.
2. Edberg SC, Samuels S. Rapid colormetric test for the determination of hippurate hydrolysis of group B *Streptococcus*. *J Clin Micro* 1976; 3:49-50.
3. Murray PR, Baron E, Pfaller M, Tenover F, Tenover F, Tenover R. *Manual of clinical microbiology*. 7th ed. Washington: ASM, 1999.
4. MacFaddin JF. *Biochemical tests for the identification of medical bacteria*. 3rd ed. Philadelphia: Lippincott Williams & Wilkins, 2000.

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